

CLAIMS

1. A polynucleotide sequence which is a polynucleotide sequence for a target gene comprising an isolated or purified single strand polynucleotide sequence comprising continuous components (I) + (II) + (III) wherein

the polynucleotide sequence for a target gene has an RNA function suppression activity in relation to RNA having a sequence complementary to either the component (I) or (III) or a partial sequence thereof,

where, the component (III) comprises a continuous polynucleotide sequence of 15 to 30 that has a sequence complementary to the target gene,

the component (II) is a nucleotide sequence or non-nucleotide sequence with a base length of from 0 base to 10 kilobases (where, 0 base means a bond), and

the component (I) is a polynucleotide sequence comprising a sequence complementary to the component (III).

2. The polynucleotide sequence according to claim 1 wherein the polynucleotide sequence of the component (III) comprises DNA or RNA.

3. The polynucleotide sequence according to claim 1 wherein the component (I) or (III) further has a sequence comprising from 1 to several U, T, G, C, or A bases on at least one terminal, or has deleted, substituted or added inside of the complementary sequence.

4. The polynucleotide sequence according to claim 1 wherein the polynucleotide sequence is obtained by chemical synthesis or gene recombination technology.

5. A polynucleotide sequence for a target gene comprising single strand RNA of SEQ ID No. 1 or 2.

6. The polynucleotide sequence according to claim 1 wherein the component (II) is a nucleotide sequence or a non-nucleotide sequence, or a combination thereof.

7. The polynucleotide sequence according to claim 6 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of 1 base or more and less than 10 kilobases.

8. The polynucleotide sequence according to claim 7 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of a length of from 1 base to several hundred bases.

9. The polynucleotide sequence according to claim 8 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of a length of from 1 base to several dozen bases.

10. The polynucleotide sequence according to claim 9 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of a length of from 1 base to 20 bases.

11. The polynucleotide sequence according to claim 10 wherein the component (II) is indicated by SEQ ID No. 3 or 4.

12. The polynucleotide sequence according to claim 1 wherein the nucleotide sequence or non-nucleotide sequence of

the component (II) comprises PNA, a cytoplasm translocation sequence, a sequence having a decoy activity, an interferon induction suppressing sequence, a sequence having any of RNase suppression activity, antisense activity, ribozyme activity, or transfer RNA, or a combination of these.

13. A method for manufacturing the polynucleotide sequence of any of claims 1 to 12 by chemical synthesis or gene recombination technology.

14. A recombinant vector wherein the polynucleotide sequence for a target gene of any of claims 1 to 12 is inserted in a vector.

15. A method of manufacturing the recombinant vector of claim 14 wherein the polynucleotide sequence for a target gene of any of claims 1 to 12 is inserted in a vector.

16. A method for screening pharmaceutical product target genes using the polynucleotide sequence for a target gene of any of claims 1 to 12, which is a screening method for assaying compounds to stimulate or suppress functions related to a target gene by introducing an isolated or purified single strand polynucleotide sequence comprising continuous components (I) + (II) + (III) in cells or tissues, and using a single strand polynucleotide sequence to increase or decrease the RNA function suppression activity of genes having a sequence complementary to the polynucleotide sequences of either of the component (I) or (III); wherein the method for screening pharmaceutical product target genes employs any one method selected from the following methods:

(a) using labeling directly or indirectly bonded to a candidate compound to measure the binding of the candidate compound and a polypeptide of an amino acid sequence that is coded by the target gene, or a target gene expression product (or a cell or membrane thereof that carries the polypeptide of an amino acid sequence that is coded by the target gene, or a target gene expression product), or a fusion protein thereof;

(b) measuring in the presence of a labeled competition substance the binding of a candidate compound and a cell into which the single strand polypeptide sequence has been introduced (or cells or the membrane thereof carrying the single strand polypeptide sequence), or a fusion substance thereof;

(c) using a detection system applied to a cell or cell membrane carrying a polypeptide of an amino acid sequence that is coded by the target gene or an expression product of the target gene to determine whether or not a candidate compound has a signal produced by suppressing or activating the polypeptide or expression product of the target gene based on the single strand polynucleotide sequence;

(d) preparing a mixture by simultaneously mixing a candidate substance and a solution containing an amino acid sequence that is coded by the target gene or an expression product of the target gene, measuring the activity of the polypeptide or the expression product of the target gene in the mixture, and comparing the activity of the mixture with that of a standard; and

(e) detecting the effect in the cell that the candidate compound has on the mRNA that codes the polypeptide of the amino acid sequence that is coded by the target gene, and on the product of the polypeptide of the amino acid sequence coded by the target gene.

17. A pharmaceutical composition taking the polynucleotide sequence for a target gene according to any of claims 1 to 12 as the active ingredient.

18. A pharmaceutical composition taking the recombinant vector of claim 14 as the active ingredient.

19. A method for introducing an isolated or purified single strand polynucleotide sequence comprising continuous components (I) + (II) + (III) into cells or tissues, and to suppress the function of a target gene based on an RNA function suppression activity of a gene having a sequence complementary to the polynucleotide sequence of either of the component (I) or (III),

where, the component (III) comprises a continuous polynucleotide sequence of 15 to 30 that has a sequence complementary to the target gene,

the component (II) is a nucleotide sequence or non-nucleotide sequence with a base length of from 0 base to 10 kilobases (where, 0 base means a bond), and

the component (I) is a polynucleotide sequence comprising a sequence complementary to the component (III).

20. The method according to claim 19 wherein the nucleotide sequence comprising polynucleotides of the component (III) comprises DNA or RNA.

21. The method according to claim 19 wherein the component (I) or (III) is DNA or RNA that has a sequence comprising from 1 to several of U, T, G, C, or A bases on any terminal, or has deleted, substituted or added inside.

22. The method according to claim 19 wherein the polynucleotide sequence is obtained by chemical synthesis or gene recombination technology.

23. The method according to claim 19 wherein the single strand polynucleotide sequence comprises a single strand RNA of SEQ ID No. 1 or 2.

24. The method according to claim 19 wherein the component (II) is a nucleotide sequence or a non-nucleotide sequence, or a combination thereof.

25. The method according to claim 24 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of 1 base or more and less than 10 kilobases.

26. The method according to claim 25 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of a length of from 1 base to several hundred bases.

27. The method according to claim 26 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of a length of from 1 base to several dozen bases.

28. The method according to claim 27 wherein the nucleotide sequence of the component (II) comprises a nucleotide sequence of a length of from 1 base to 20 bases.

29. The method according to claim 28 wherein the component (II) is indicated in SEQ ID No. 3 or 4.

30. The method according to claim 18 wherein the nucleotide sequence or non-nucleotide sequence of the component (II) comprises PNA, a cytoplasm translocation sequence, a sequence having a decoy activity, an interferon induction suppressing sequence, a sequence having any of RNase suppression activity, antisense activity, ribozyme activity, or transfer RNA, or a combination of these.

31. A method for suppressing expression of the protein of a target gene based on the method for suppressing the function of a target gene according to claims 19 to 30.

32. A method for suppressing the activity of a transcript of a target gene based on the method for suppressing the function of a target gene according to claims 19 to 30.

33. A knockdown cell or tissue or a non-human knockdown animal or a knockdown plant produced and cultured by the method of claim 31 or 32.

34. A knockdown cell or tissue or a non-human knockdown animal according to claim 33 that is for organ transplants.

35. A gene therapy agent comprising a pharmaceutical composition according to claim 17 or 18.

36. A method for testing the function of a target gene by introducing an isolated or purified single strand polynucleotide

sequence comprising continuous components (I) + (II) + (III) into cells, tissues, non-human animals, or plants to have an RNA function suppression activity of a gene having a sequence complementary to the polynucleotide sequence of either of the component (I) or (II),

where, the component (III) comprises a continuous polynucleotide sequence of 15 to 30 that has a sequence complementary to the target gene,

the component (II) is a nucleotide sequence or non-nucleotide sequence with a base length of from 0 base to 10 kilobases (where, 0 base means a bond), and

the component (I) is a polynucleotide sequence comprising a sequence complementary to the component (III).

37. A method for detecting a candidate compound to reinforce the function of a target gene comprising the steps of:

introducing an isolated or purified single strand polynucleotide sequence comprising continuous components (I) + (II) + (III) into cells, tissues, non-human animals, or plants after culturing the test compound together with the cells, tissues, non-human animals, or plants; and comparing to a control the RNA function suppression activity of the RNA of a gene having a sequence complementary to the polynucleotide sequence of either of the component (I) or (III),

where, the component (III) comprises a continuous polynucleotide sequence of 15 to 30 that has a sequence complementary to the target gene,

the component (II) is a nucleotide sequence or non-nucleotide sequence with a base length of from 0 bases to 10 kilobases (where, 0 bases means a bond), and

the component (I) is a polynucleotide sequence comprising a sequence complementary to the component (III).

38. A polynucleotide sequence for a target gene according to any of claims 1 to 4 wherein the component (III) comprises any type of 1 to 5 ribonucleotides continuing at the 18 to 25 ribonucleotides complementary to the target gene, and the component (I) comprises 18 to 25 ribonucleotides complementary to the 18 to 25 nucleotides of the component (III).

39. A method for synthesizing nucleotides for target genes including the following steps:

(i) preparing a single strand nucleotide comprising component (I) and (II) such that several nucleotides of the 3' terminal of component (II) are complementary to several nucleotides of component (I) or (II);

(ii) synthesizing component (III) based on nucleotide synthesis enzyme activity using this single strand nucleotide comprising components (I) and (II), or introducing this single strand nucleotide comprising components (I) and (II) into a cell and synthesizing component (III) based on the nucleotide synthesis enzyme activity present inside the cell.

40. A nucleotide for a randomized target gene obtained by the method of claim 39, wherein the components (I) and (III) are random oligonucleotides.